

REPORT DOCUMENTATION PAGE

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OMB No. 0704-0188

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1. AGENCY USE ONLY (Leave blank)		2. REPORT DATE 2007	3. REPORT TYPE AND DATES COVERED Journal Article-J Appl. Physiol.	
4. TITLE AND SUBTITLE Rehydration with Fluid of Varying Tonicities: Effects on Fluid Regulatory Hormones and Exercise Performance in the Heat			5. FUNDING NUMBERS	
6. AUTHOR(S) R.W. Kenefick, C.M. Maresh, L.E. Armstrong, D. Riebe, M.E. Echegaray, J.W. Castellani				
7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES) Thermal and Mountain Medicine Division U.S. Army Research Institute of Environmental Medicine Natick, MA 01760-5007			8. PERFORMING ORGANIZATION REPORT NUMBER M06-36	
9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES) Same as #7 above			10. SPONSORING / MONITORING AGENCY REPORT NUMBER	
11. SUPPLEMENTARY NOTES				
12a. DISTRIBUTION / AVAILABILITY STATEMENT Approved for public release; distribution unlimited			12b. DISTRIBUTION CODE	
13. ABSTRACT (Maximum 200 words) This study examined the effects of rehydration (Rehy) with fluids of varying tonicities and routes of administration after exercise-induced hypohydration on exercise performance, fluid regulatory hormone responses, and cardiovascular and thermoregulatory strain during subsequent exercise in the heat. On four occasions, eight men performed an exercise-dehydration protocol of ~185 min (33°C) to establish a 4% reduction in body weight. Following dehydration, 2% of the fluid lost was replaced during the first 45 min of a 100-min rest period by one of three random Rehy treatments (0.9% saline intravenous; 0.45% saline intravenous; 0.45% saline oral) or no Rehy (no fluid) treatment. Subjects then stood for 20 min at 36°C and then walked at 50% maximal oxygen consumption for 90 min. Subsequent to dehydration, plasma Na ⁺ , osmolality, aldosterone, and arginine vasopressin concentrations were elevated ($P < 0.05$) in each trial, accompanied by a -4% hemoconcentration. Following Rehy, there were no differences ($P > 0.05$) in fluid volume restored, post-rehydration (Post-Rehy) body weight, or urine volume. Percent change in plasma volume was 5% above pre-Rehy values, and plasma Na ⁺ , osmolality, and fluid regulatory hormones were lower compared with no fluid. During exercise, skin and core temperatures, heart rate, and exercise time were not different ($P > 0.05$) among the Rehy treatments. Plasma osmolality, Na ⁺ , percent change in plasma volume, and fluid regulatory hormones responded similarly among all Rehy treatments. Neither a fluid of greater tonicity nor the route of administration resulted in a more rapid or greater fluid retention, nor did it enhance heat tolerance or diminish physiological strain during subsequent exercise in the heat.				
14. SUBJECT TERMS fluid regulation; hydration state; dehydration; environment; osmotic load			15. NUMBER OF PAGES 7	
			16. PRICE CODE	
17. SECURITY CLASSIFICATION OF REPORT Unclassified	18. SECURITY CLASSIFICATION OF THIS PAGE Unclassified	19. SECURITY CLASSIFICATION OF ABSTRACT Unclassified	20. LIMITATION OF ABSTRACT Unclassified	

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J Appl Physiol 102:1899-1905, 2007. First published Feb 22, 2007; doi:10.1152/japplphysiol.00920.2006

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Rehydration with fluid of varying tonicities: effects on fluid regulatory hormones and exercise performance in the heat

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Submitted 21 August 2006; accepted in final form 15 February 2007

Kenefick RW, Maresh CM, Armstrong LE, Riebe D, Echegaray ME, Castellani JW. Rehydration with fluid of varying tonicities: effects on fluid regulatory hormones and exercise performance in the heat. *J Appl Physiol* 102: 1899–1905, 2007. First published February 22, 2007; doi:10.1152/jappphysiol.00920.2006.—This study examined the effects of rehydration (Rehy) with fluids of varying tonicities and routes of administration after exercise-induced hypohydration on exercise performance, fluid regulatory hormone responses, and cardiovascular and thermoregulatory strain during subsequent exercise in the heat. On four occasions, eight men performed an exercise-dehydration protocol of ~185 min (33°C) to establish a 4% reduction in body weight. Following dehydration, 2% of the fluid lost was replaced during the first 45 min of a 100-min rest period by one of three random Rehy treatments (0.9% saline intravenous; 0.45% saline intravenous; 0.45% saline oral) or no Rehy (no fluid) treatment. Subjects then stood for 20 min at 36°C and then walked at 50% maximal oxygen consumption for 90 min. Subsequent to dehydration, plasma Na⁺, osmolality, aldosterone, and arginine vasopressin concentrations were elevated ($P < 0.05$) in each trial, accompanied by a ~4% hemoconcentration. Following Rehy, there were no differences ($P > 0.05$) in fluid volume restored, post-rehydration (Post-Rehy) body weight, or urine volume. Percent change in plasma volume was 5% above pre-Rehy values, and plasma Na⁺, osmolality, and fluid regulatory hormones were lower compared with no fluid. During exercise, skin and core temperatures, heart rate, and exercise time were not different ($P > 0.05$) among the Rehy treatments. Plasma osmolality, Na⁺, percent change in plasma volume, and fluid regulatory hormones responded similarly among all Rehy treatments. Neither a fluid of greater tonicity nor the route of administration resulted in a more rapid or greater fluid retention, nor did it enhance heat tolerance or diminish physiological strain during subsequent exercise in the heat.

fluid regulation; hydration state; dehydration; environment; osmotic load

HYPHYDRATION INDUCED BY exercise-heat stress decreases plasma volume (PV) and increases plasma sodium concentration ([Na⁺]) and osmolality (Osm) (4, 23, 26). Concomitantly, exercise following hypohydration raises plasma concentrations of arginine vasopressin ([AVP]) (2, 7, 15) and aldosterone ([ALD]) (2, 8, 12) relative to euhydrated control conditions. However, it is not evident whether the lower PV or higher [Na⁺] caused by hypohydration influenced the exercise-induced changes in [AVP] and [ALD]. Furthermore, rehydration (Rehy) during or following exercise generally lowers

[AVP] and [ALD] (2, 5, 17, 28), but the mechanisms for this are unclear.

Several investigations have addressed the roles of PV, [Na⁺], and Osm during Rehy on fluid-regulating hormones. Moses et al. (18) used dehydration (Dehy) and intravenous (IV) infusion of 5% saline to examine mediators of AVP release and suggested that PV expansion following infusion may serve to attenuate the osmotic stimulus for AVP secretion. This finding implies that AVP would remain the same if IV fluids of varying tonicities were used during Rehy, if PV was restored similarly. Nose et al. (21) reported that, during recovery from Dehy, rehydrating orally with a Na⁺-containing drink (compared with drinking water) better restored PV and lowered plasma renin activity and ALD to a greater extent, suggesting PV changes have a larger role than osmotic changes in mediating renin and ALD responses after oral Rehy (21). They stated that an inferior Rehy occurs when drinking hypotonic water, because it removes the osmotic drive for drinking and increases free water clearance, suggesting that Rehy with fluids of greater tonicity would better maintain PV compared with fluids of lesser tonicity. It is unknown whether tonicity differences would also cause differential PV and ALD responses after exercise-induced Dehy, if Rehy fluid were delivered via IV, rather than orally.

In addition, whether IV Rehy following exercise-induced hypohydration should be isosmotic or hypotonic has practical implications, if multiple bouts of exercise in the heat are performed and if the type of IV Rehy solution is found to differentially affect PV restoration or plasma [Na⁺] and Osm. Numerous studies have shown that either lower PV or higher Osm causes the sweating rate or cutaneous blood flow (9) to be lower at any given core temperature and for these thermoregulatory effector responses to begin at a higher core temperature (22). Changes in these thermoregulatory responses subsequent to different IV treatments could potentially affect core temperature and exercise performance during repeated exercise bouts.

The primary purpose of this study was to examine the effects of hypovolemia and [Na⁺] on fluid-regulatory hormone responses to exercise in the heat, subsequent to exercise-induced hypohydration. To test the hypothesis that differences in either PV or [Na⁺] would affect plasma [AVP] and [ALD] during exercise heat stress, measurements were made following Dehy with two IV treatments (0.9 and 0.45% NaCl), an oral Rehy treatment (0.45% Oral), and a no-fluid control treatment (NF). We hypothesized that, following Dehy: 1) IV Rehy with a fluid

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of greater tonicity (0.9% saline) would increase the plasma $[Na^+]$ above that of a fluid of lesser tonicity (0.45% saline); 2) the greater $[Na^+]$ associated with the 0.9% IV treatment would subsequently elevate plasma Osm and stimulate AVP secretion above that of the 0.45% IV or Oral treatments; 3) due to the more rapid restoration of PV in the IV treatment and the greater fluid retention in the 0.9% IV treatment (a result of higher [AVP]), there will be less cardiovascular and thermoregulatory strain, greater heat tolerance, and exercise duration during subsequent exercise in the heat, compared with the 0.45% Oral and IV saline treatments. A secondary focus was to determine whether the route of fluid administration (Oral vs. IV) would result in any physiological differences pertaining to fluid-regulatory responses.

METHODS

Subjects. Eight men, unacclimatized to heat, volunteered to participate in this investigation. Physical characteristics (mean \pm SE) were as follows: age, 22.1 ± 0.8 yr; height, 179.6 ± 1.5 cm; weight, 73.6 ± 2.4 kg; maximal oxygen consumption ($\dot{V}O_{2\max}$), 57.9 ± 1.6 ml \cdot kg $^{-1}\cdot$ min $^{-1}$; percent body fat, $7.7 \pm 0.9\%$. Before participation, each subject completed a written, informed consent document and a medical history questionnaire after being informed of the purpose of the experiment and possible risks. The study protocol was approved in advance by the Committee on the Use of Human Subjects in Research at the University. Subjects were paid for their participation.

Preliminary measures. $\dot{V}O_{2\max}$ was determined using a continuous treadmill running test, as previously described (9). Briefly, the $\dot{V}O_{2\max}$ test was preceded by a 7-min warm-up run at ~ 160 – 200 m/min, 0% grade. During the maximal oxygen test, speed was kept constant for 160–220 m/min, 0% grade, for the first 4 min, after which the grade was raised to 4%. Thereafter, the grade was increased by 2% every 2 min until volitional exhaustion. The hydrostatic weighing technique described by Katch and Katch (14) was utilized to determine body density. Body fat was calculated from the formula of Brozek et al. (3). Measurement of residual lung volume was made using a nitrogen washout technique (29) on a pulmonary analysis system (model 1070, Medical Graphics, St. Paul, MN).

Experimental design. Testing involved three treatments, each consisting of three stages, as follows: 1) exercise-induced Dehy, 2) Rehy, and 3) moderate exercise in the heat. The protocols were randomly assigned and separated by at least 14 days. The Rehy treatments were as follows: 0.9% IV infusion (0.9% IV); 0.45% IV infusion (0.45% IV); 0.45% Oral Rehy (0.45% Oral), and no Rehy (NF). Subjects were asked to consume similar diets during the 3 days before each experimental trial, verified by a 3-day dietary record. These food records were then analyzed for kilocalories, carbohydrate, fat, protein, sodium, and potassium content (Food Processor II, ESHA Research, Salem, OR). Subjects were asked to refrain from any recreational activities or exercise training for 24 h before experimental testing. They were also instructed to drink 450 ml of water the night before testing, and 450 ml of water the morning of testing, and to abstain from eating for 12 h before each experimental treatment.

Experimental treatments. On arrival at the laboratory (0700), subjects provided a urine sample for determination of urine-

specific gravity (U_{sg} ; Spartan Refractometer, model A 300 CL). A U_{sg} of 1.023 ± 0.006 (1) was used to verify that the subject was adequately hydrated before each trial. Hydration status was additionally verified by a pre-Dehy plasma Osm value of $\leq 286 \pm 3$ mosmol/kgH $_2$ O (1). Subjects were then fitted with a heart rate monitor (UNIQ heartwatch, Computer Instrument, Hempstead, NY), and a flexible thermistor (Yellow Springs Instruments, series 401, Yellow Springs, OH) was inserted 10 cm beyond the external anal sphincter to monitor rectal temperature (T_{re}). A Teflon catheter was then inserted into a superficial forearm vein, and a male luer adapter (model 5877, Abbott Hospital, Chicago, IL) was inserted into the catheter port for acquisition of subsequent blood samples. The catheter port and male luer adapter were kept patent with heparin Lock Flush Solution. The subject then entered the environmental chamber (model 2000, Minus-Eleven, Malden, MA) and stood quietly during a 20-min equilibration period. A 26-ml blood sample (baseline) was taken, and subjects then consumed a standard breakfast of one bagel, one banana, and 240–350 ml (depending on body weight) of fruit juice.

Immediately before each of the four experimental trials, subjects performed a Dehy protocol in 33°C air, to reduce body weight by $\sim 4\%$. The Dehy protocol consisted of alternating stationary cycling (117 ± 9 W; model 818E, Monark, Sweden) and treadmill walking (1.6 ± 0.1 m/s; $5 \pm 1\%$ grade; Quinton, Seattle, WA) at a 25-to-5-min ratio (exercise/rest) for each modality. Body weight was measured during each rest interval. Urine was collected throughout the Dehy period and was included as part of the weight loss. Subjects continued exercising until the desired weight loss was achieved. The last exercise mode before the 4% weight loss was always walking, to ensure an upright posture. The time to achieve the 4% decrease in body weight before each of the three treatments was consistent for each individual subject (Table 1). The mean exercise intensity for the four Dehy trials ranged from 49.8 to 51.1% of $\dot{V}O_{2\max}$. The mean ambient temperature and percent relative humidity were $33.0 \pm 0.1^\circ\text{C}$ and $47.6 \pm 0.5\%$, respectively. Airflow (2.3 m/s) was generated by a fan directed at the subject.

Following the exercise-induced Dehy protocol, a 26-ml blood sample was taken, and the subject exited the environmental chamber to a $25.5 \pm 0.2^\circ\text{C}$ environment. Subjects assumed a recumbent position, and after a 15-min rest period received one of the three Rehy treatments or NF control over a 45-min period. An experienced IV nurse, using a butterfly

Table 1. Selected Dehy and Rehy variables

	0.45% IV	0.45% Oral	0.9% IV	NF
Dehy % $\dot{V}O_{2\max}$, ml \cdot kg $^{-1}\cdot$ min $^{-1}$	51 ± 2	50 ± 2	50 ± 3	51 ± 2
Exercise time, min	190 ± 12	182 ± 8	179 ± 12	180 ± 10
Post-Dehy urine volume, liter	0.34 ± 0.12	0.40 ± 0.10	0.44 ± 0.11	0.48 ± 0.09
Post-Rehy urine volume, liter	0.06 ± 0.02	0.08 ± 0.02	0.08 ± 0.02	0.08 ± 0.02
Post-Dehy, %weight loss	4.4 ± 0.07	5.1 ± 0.1	4.5 ± 0.07	4.6 ± 0.07
Post-Rehy %weight loss	2.1 ± 0.10	2.3 ± 0.1	2.1 ± 0.10	$4.6 \pm 0.04^*$

Values are means \pm SE; $n = 8$. IV, intravenous; Oral, oral rehydration; NF, no fluid control; Dehy, dehydration; $\dot{V}O_{2\max}$, maximal oxygen consumption; Post-Dehy, post-dehydration; Post-Rehy, post-rehydration. *Difference from 0.9% IV, 0.45% IV, and 0.45% Oral treatments ($P < 0.05$).

catheter (1.5 in., 16 gauge; Abbot Laboratories, North Chicago, IL), administered the IV infusions in the arm opposite to the indwelling venous cannula. During the NF trials, a cannula was not placed in the opposite arm during the Rehy period. The rate of IV infusion was $0.56 \text{ ml} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$. Oral Rehy consisted of 4 g of Sugar Free Tropical Punch Kool-Aid dissolved in 889 ml of 0.45% NaCl and 111 ml of distilled, deionized water chilled to 4°C for palatability. The composition of 0.45% Oral fluid was $78.6 \pm 1.0 \text{ meq/l Na}^+$, $0.96 \pm 0.02 \text{ meq/l K}^+$, and $2.54 \pm 0.07 \text{ meq/l Ca}^{2+}$. The Osm was $145.6 \pm 1.1 \text{ mosmol/kgH}_2\text{O}$. Fluid given in the 0.45% Oral trial was weighed before each drink and was given every 5 min over a 45-min period. The fluid volumes given were not different ($P > 0.05$) and were $1,890 \pm 45$, $1,856 \pm 65$, and $1,889 \pm 62 \text{ ml}$ for the 0.45% IV, 0.45% Oral, and 0.9% IV treatments, respectively. The rate of IV infusion and oral Rehy over 45 min was chosen to allow for a fluid volume restoration to be similar to the upper range for orally ingested fluids after exercise-induced Dehy (20). Subjects then stood for 55 min in the laboratory to allow for equilibration of the fluid and then reentered the environmental chamber.

Subjects reentered the climatic chamber, equilibrated (standing for 20 min), and a 26-ml blood draw was performed. Subjects then consumed 1 g/kg body wt of a commercial carbohydrate product (Skittles, M and M Mars, Hackettstown, NJ) and 100 ml of distilled, deionized water. Carbohydrates were given before the second exercise bout to offset the possible loss of muscle glycogen during the Dehy protocol. Subjects then performed a 90-min exercise bout of treadmill walking, up a 3–5% grade at $\sim 50\%$ of $\dot{V}\text{O}_{2\text{max}}$. The exercise bout could be stopped either by subjects (volitional exhaustion), or by the researchers, due to heart rate ($>180 \text{ beats/min}$ for 5 consecutive min) or T_{re} ($>39.5^\circ\text{C}$) criteria or symptoms of heat exhaustion. Walking speed was verified for each test with a hand-held tachometer (model 8204–20, Cole-Parmer Instrument, Chicago, IL). A 26-ml blood draw was performed at the completion of the exercise bout. The mean temperature and relative humidity were $35.9 \pm 0.1^\circ\text{C}$ and $46.6 \pm 2\%$, respectively, with airflow at 2.3 m/s .

Physiological measures. During all Dehy bouts, the subjects' heart rate and T_{re} were measured every 15 min to monitor physiological strain. Oxygen consumption was measured once every 30 min during the Dehy protocol for a 7- to 10-min period.

Expired gas samples were analyzed in each breath (Medical Graphics CPX-D system, Medical Graphics, St. Paul, MN). During exercise in the heat, thermocouples (series 400, Yellow Springs Instruments) secured on the chest, arm, thigh, and calf were used to measure skin temperature (T_{sk}), computed from a weighted mean of the four local skin measurements (20).

Analysis of blood samples. Blood measures were analyzed at four time points: pre-dehydration (Pre-Dehy), post-dehydration (Post-Dehy), preexercise in the heat (Pre-Ex/Post-Rehy), and postexercise in the heat (Post-Ex). Blood was transferred from sterile syringes to tubes containing EDTA, lithium heparin, or SST gel and clot activator for serum separation, depending on the analytical specifications for each blood variable. Tubes were centrifuged (Marathon 12KBR Refrigerated Centrifuge, Fisher Scientific, Pittsburgh, PA) for 15 min using 760 g at 4°C . Serum or plasma was separated from each sample and stored at -80°C for later analysis. Samples of whole blood

were taken for analysis of hemoglobin (Hb) and hematocrit (Hct). Following centrifugation, plasma was separated and analyzed for Osm and Na^+ . Absolute changes in plasma AVP, ALD, Na^+ , Osm, and percent change of PV (% ΔPV) were calculated and analyzed relative to the Pre-Dehy time point.

Hct was determined in triplicate by the microcapillary technique following centrifugation for 4 min at $9,500 \text{ g}$. Values were not corrected for trapped plasma. Hb was determined in triplicate by the cyanomethemoglobin method (kit 525, Sigma Chemical, St. Louis, MO). % ΔPV was calculated using the equation of Dill and Costill (9a) from appropriate Hct and Hb values. All % ΔPV values were calculated using Post-Dehy as the initial time point. Plasma Osm was measured in triplicate via freezing point depression (MicroOsmometer model 3MO, Advanced Instruments, Needham Heights, MA). Plasma and urine $[\text{Na}^+]$ were determined in duplicate by selective ion-sensitive electrodes (model 984-S AVL Scientific, Roswell, GA).

Endocrine measures. After extraction on silica columns (DiaSorin, Stillwater, MN), plasma AVP was determined in duplicate by a commercially available radioimmunoassay kit (DiaSorin). AVP values were not corrected for extraction recovery, which was 91.3%. The limit of the detection for this assay was $\sim 1.4 \text{ pg/ml}$. Serum levels of ALD were determined in duplicate by radioimmunoassay (Diagnostics Products, Los Angeles, CA). Assay sensitivity was 11.0 pg/ml . Within- and between-assay coefficients of variability for both assays were $<5\%$. [AVP] and [ALD] were not corrected for % ΔPV .

Statistical analysis. A 4×3 (treatment \times time) repeated-measures ANOVA was used to compare differences among trials. A Newman-Keuls post hoc analysis was employed to determine significant differences within and between conditions. The 0.05 level of significance was selected. Studies examining fluid-regulating hormones during Dehy have a coefficient of variation of 5%. A power analysis selecting conventional α ($P < 0.05$) and β (0.20) parameters showed that eight subjects would provide sufficient power to detect an effect greater than or equal to the anticipated coefficient of variation. All data are presented as means \pm SE.

RESULTS

Dehy. Pre-Dehy dietary carbohydrate, protein, fat, sodium, and total kilocalorie intakes were similar over the 3 days before each experimental treatment. Pre-Dehy body weights were not different ($P > 0.05$) and were 75 ± 2 , 74.3 ± 2.6 , 74 ± 3 , and $74 \pm 3 \text{ kg}$ for the 0.45% IV, 0.45 oral, 0.9% IV, and NF treatments, respectively. The exercise intensity during the Dehy phase across the four experimental conditions ranged from 49.8 to 51.1% of $\dot{V}\text{O}_{2\text{max}}$. There were no differences ($P > 0.05$) among the three treatments and the NF trial for Dehy time, percent weight loss, urine volume, and exercise intensity (% $\dot{V}\text{O}_{2\text{max}}$) during Dehy (Table 1).

Pre- and Post-Dehy plasma and urine values. Pre-Dehy plasma AVP, ALD, Osm, and $[\text{Na}^+]$ were not different ($P > 0.05$) among treatments. In addition, Pre-Dehy urine Osm, $[\text{Na}^+]$, and U_{sg} values were not different ($P > 0.05$) among treatments. Following Dehy, plasma values of AVP, ALD, Osm, and $[\text{Na}^+]$ were elevated ($P < 0.05$) above Pre-Dehy values but were not different among the Rehy treatments and NF trial Post-Dehy. Post-Dehy urine Osm, $[\text{Na}^+]$, and U_{sg} were also not different ($P > 0.05$) from Pre-Dehy values

(Table 2). In addition, the $\% \Delta PV$ calculated from the Pre-Dehy was not different ($P > 0.05$) among the Rehy treatments and the NF trial; the change from Pre- to Post-Dehy averaged -5.2% for the Rehy treatments and the NF trial (see Fig. 2A).

Rehy and exercise. There were no differences in the volume of IV or Oral fluid (0.9% saline IV, 0.45% saline IV, and 0.45% saline Oral) given during the Rehy period (Table 1). Post-Rehy urine volume was similar among the Rehy and NF trials. The percentage of weight loss Post-Rehy (compared with the Pre-Dehy body weight) was similar among the 0.45% IV, 0.45 Oral, and 0.9% IV treatments and were significantly lower ($P < 0.05$) than in the NF trial (Table 1).

The absolute change from Pre-Dehy in plasma $[Na^+]$ was lower ($P < 0.05$) in the 0.45% Oral treatment compared with both the 0.9% IV and NF treatments after Rehy (Pre-Ex time point). However, Post-Ex in the heat, there were no differences ($P > 0.05$) in the absolute change in Na^+ among any of the treatments or the NF trial (Fig. 1A). The absolute change in plasma Osm in the NF trial was greater than that observed in the 0.45% IV and Oral treatments following Rehy (Pre-Ex time point); however, following exercise in the heat, none of the Rehy treatments or the NF trial were different ($P > 0.05$) (Fig. 1B).

Following Rehy, at the Pre-Ex and Post-Ex time points, the absolute change in plasma AVP was greater ($P > 0.05$) in the NF trial compared with the Rehy treatments. Following exercise in the heat, there was an increase in the absolute plasma AVP in all three treatments and the NF trial, but this change was only different ($P < 0.05$) from Pre-Ex in the NF trial (Fig. 1C).

Subsequent to Rehy, the $\% \Delta PV$ at the Pre-Ex time point was not different ($P > 0.05$) among the Rehy treatments, but was greater ($P < 0.05$) than in the NF trial. Following exercise in the heat, there was a significant hemoconcentration in the Rehy treatments ($P < 0.05$) relative to the Pre-Ex time point. In the NF trial, the $\% \Delta PV$ did decrease slightly Post-Ex; however, there was no difference ($P > 0.05$) in the $\% \Delta PV$ Post-Ex among the NF trial and Rehy treatments (Fig. 2A).

At the Pre-Ex and Post-Ex time points, the absolute change in plasma ALD was greater ($P > 0.05$) in the NF trial compared with the Rehy treatments. Following exercise in the heat, there was an increase ($P < 0.05$) in the absolute change in plasma ALD in all three treatments and the NF trial (Fig. 2B).

Selected Pre- and Post-Ex thermoregulatory and exercise performance time. Exercise performance times were longer ($P < 0.05$) in 0.45% IV, 0.45% Oral, and 0.9% IV treatments vs. NF (Table 3). Exercise trials in the heat were terminated for the following reasons: during the 0.45% IV, three subjects reached $39.5^\circ C$, one reached the heart rate limit of >180 beats/min for 5 consecutive min, one subject developed signs and symptoms of heat exhaustion, and three completed 90 min of exercise; during the 0.45% Oral trial, two subjects reached $39.5^\circ C$, two subjects exhibited symptoms of fatigue, and four completed 90 min of exercise; during the 0.9% IV trial, two subjects reached $39.5^\circ C$, two subjects exhibited symptoms of fatigue, and four completed 90 min of exercise; during the NF trial two subjects reached $39.5^\circ C$, four subjects exhibited symptoms of fatigue, and two completed 90 min of exercise.

Pre-Ex measures of T_{re} were lower ($P < 0.05$) in the Rehy treatments compared with the NF trial. Immediate Post-Ex, T_{re} were higher ($P < 0.05$) than Pre-Ex temperatures for all Rehy treatments and the NF trial, but were not different ($P > 0.05$) among each other. Pre-Ex T_{sk} were also lower ($P < 0.05$) in the Rehy treatments compared with NF; however, after exercise, T_{sk} were not different among the 0.9% IV, 0.45% IV, and Oral treatments and the NF trial. NF Pre-Ex heart rates were greater ($P < 0.05$) compared with the Rehy treatments, but Post-Ex heart rates were not different ($P > 0.05$) among treatments. All Pre-Ex Rehy treatment body weights were greater ($P < 0.05$) than NF. Only the NF Post-Ex body weights were significantly lower ($P < 0.05$) than Pre-Ex. In addition, the NF Post-Ex body weights were lower ($P < 0.05$) compared with Rehy Post-Ex values.

DISCUSSION

In the present study, a 4–5% exercise-induced hypohydration caused subjects to become hyperosmotic, hypovolemic, and elevated circulating [AVP] and [ALD] (5, 8). The results of the present investigation demonstrate that, following partial Rehy, there were no overall differences in $\% \Delta PV$ or absolute change of plasma sodium, Osm, [AVP], and [ALD] among the Rehy treatments. There was no greater fluid restoration associated with the 0.9% IV compared with the 0.45% IV treatment, nor was there a greater fluid restoration imparted by IV compared with oral Rehy. As a result, there were no cardiovascular, thermoregulatory, or performance advantages during

Table 2. Selected plasma and urine variables pre- and post-Dehy

	Pre-Dehy				Post-Dehy			
	0.45% IV	0.45% Oral	0.9% IV	NF	0.45% IV	0.45% Oral	0.9% IV	NF
Plasma								
AVP, pg/ml	5.5 ± 0.8	5.8 ± 0.9	3.8 ± 0.8	4.9 ± 1.2	$20.0 \pm 1.9^*$	$22.0 \pm 2.0^*$	$15.4 \pm 1.9^*$	$18.4 \pm 1.9^*$
ALD, pg/ml	410 ± 69	277 ± 71	386 ± 66	248 ± 62	$1,351 \pm 226^*$	$996 \pm 269^*$	$1,148 \pm 139^*$	$1,054 \pm 147^*$
Osm, mosmol/kgH ₂ O	284 ± 1	282 ± 2.0	282 ± 1	286 ± 1	$294 \pm 2^*$	$296 \pm 2^*$	$294 \pm 2^*$	$296 \pm 1^*$
Na ⁺ , meq/l	146 ± 0.4	145 ± 1.0	145 ± 1.0	146 ± 0.4	$153 \pm 1.0^*$	151 ± 1.0	$151 \pm 1.0^*$	$151 \pm 1.0^*$
Urine								
Volume (ml)					300 ± 10	500 ± 10	400 ± 10	500 ± 10
Osm, mosmol/kgH ₂ O	499 ± 117	487 ± 76	403 ± 101	432 ± 94	607 ± 108	655 ± 97	555 ± 133	617 ± 113
Na ⁺ , meq/l	61 ± 9	75 ± 14	64 ± 23	58 ± 12	44 ± 6	$24.5 \pm 9^*$	63 ± 7	52 ± 6
Urine-specific gravity	1.015 ± 0.003	1.017 ± 0.003	1.014 ± 0.003	1.010 ± 0.002				

Values are means \pm SE; $n = 8$. Pre-Dehy, pre-dehydration; AVP, arginine vasopressin; ALD, aldosterone; Osm, osmolality. *Difference from corresponding 0.45% IV, 0.45% Oral, 0.9 % IV, and NF Pre-Dehy value ($P < 0.05$).

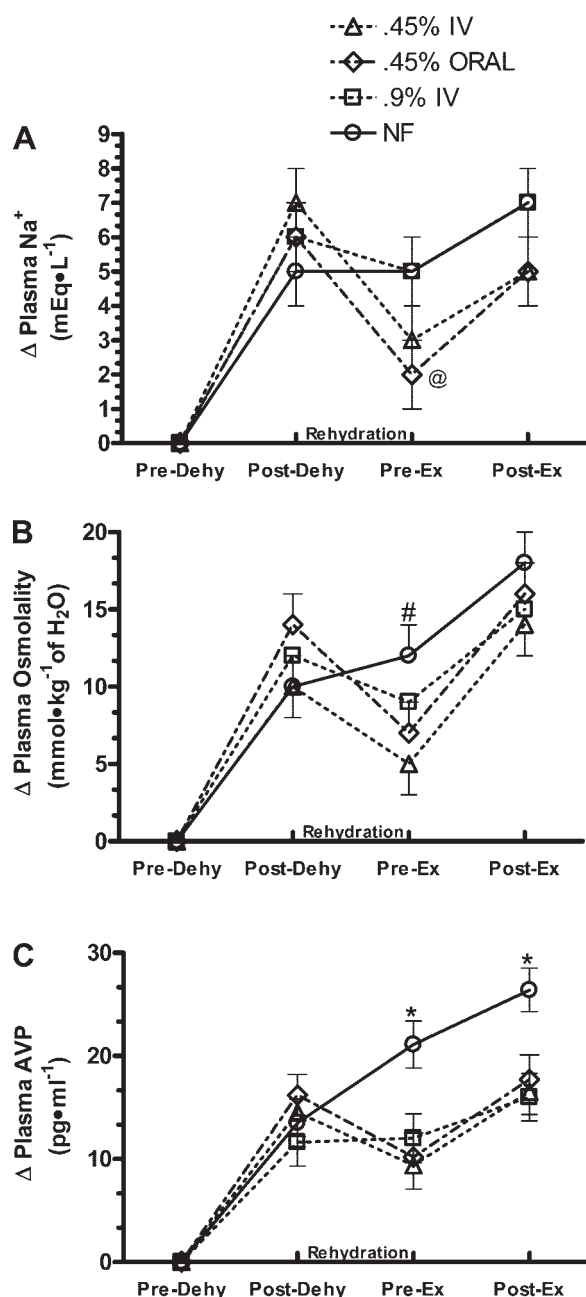


Fig. 1. Change (Δ) in plasma Na^+ (A), plasma osmolality (B), and plasma arginine vasopressin (C) as a function of time after dehydration with 0.45% intravenous (IV; Δ), 0.45% oral rehydration (Oral; \diamond), 0.9% IV (\square), and no fluid control (NF; \circ). Values are means \pm SE. Pre-dehydration (Pre-Dehy) is considered the reference point. Post-Dehy, post-dehydration; Pre-Ex, preexercise; Post-Ex, postexercise. @Significant difference ($P < 0.05$) from 0.9% IV and NF trials at the corresponding time point. #Significant difference ($P < 0.05$) from 0.45% IV and 0.45% Oral trials at the corresponding time point. *Significant difference ($P < 0.05$) from 0.9% IV, 0.45% IV, and 0.45% Oral trials at the corresponding time point.

subsequent exercise in the heat associated with either a particular IV solution or administration route.

Plasma $[\text{Na}^+]$, Osm, and AVP responses. Prolonged exercise in the heat without Rehy increases plasma [AVP] (2, 7, 16). As plasma Osm has been identified as the primary mediator of AVP release (18), we hypothesized that infusion of the 0.9% IV saline infusion would raise the plasma $[\text{Na}^+]$ and

plasma Osm above that of the 0.45% IV treatment, further stimulating AVP secretion in 0.9% IV treatment. The present results do not agree with our hypothesis. In the present study, during a 75-min post-Rehy recovery (55-min post-Rehy + 20-min equilibration), the 0.9% IV $[\text{Na}^+]$ were elevated in the 0.9% IV compared with the 0.45% IV treatment at the Pre-Ex time point, but was not significantly different, despite the greater tonicity of the 0.9% IV fluid. As a result, the absolute change in plasma Osm and [AVP] was also not different between the IV treatments. Thus the greater $[\text{Na}^+]$ in the 0.9% IV treatment may not have been large enough to cause differences in plasma Osm between the 0.9% IV and the 0.45% IV or Oral treatments.

We previously reported a reduction in plasma AVP after 55 min of Rehy with 0.9% and 0.45% IV fluid (15). In that study, plasma $[\text{Na}^+]$ in the 0.9% Rehy trial were significantly elevated above that of the 0.45% at 55 min post-Rehy. Also, while not significantly different, plasma AVP was greater after 55 min post-Rehy in the 0.9% Rehy trial compared with the 0.45% trial. Our current results (reduced [AVP] and [ALD]) agree with previous studies where subjects received water or an electrolyte solution before or during exercise in a warm environment (2, 12, 13). Follenius et al. (10) found that progressive Rehy, by either acid isotonic solution or neutral isotonic solution, equally maintained PV and blunted the AVP response. In contrast, Thompson et al. (27) reported that, in addition to increasing blood volume, infusion of hypertonic

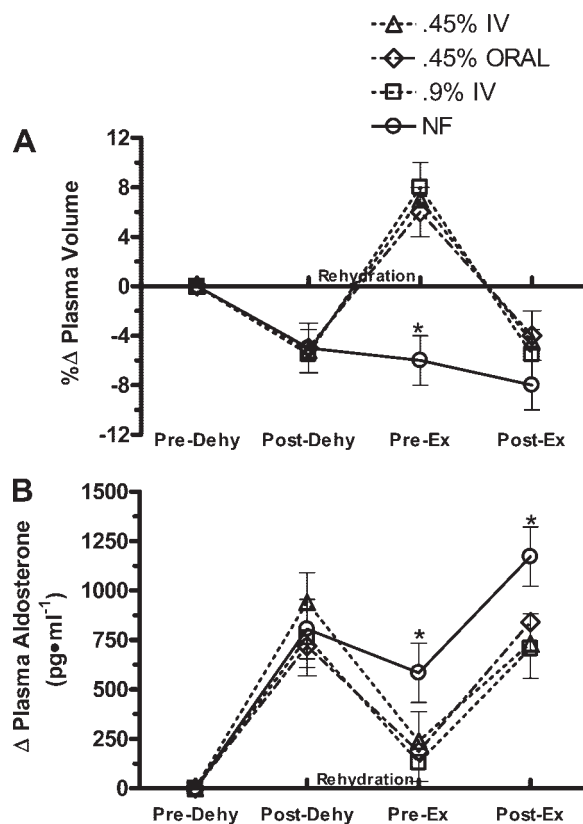


Fig. 2. Percent change in plasma volume (A) and plasma aldosterone (B) as a function of time after dehydration with 0.45% IV (Δ), 0.45% Oral (\diamond), 0.9% IV (\square), and NF (\circ). Values are means \pm SE. Pre-Dehy is considered the reference point. *Significant difference ($P < 0.05$) from 0.9% IV, 0.45% IV, and 0.45% Oral trials at the corresponding time point.

Table 3. Selected Pre-Ex and exercise performance variables

	Pre-Ex				Post-Ex			
	0.45% IV	0.45% Oral	0.9% IV	NF	0.45% IV	0.45% Oral	0.9% IV	NF
Exercise time, min					77±5	84±2	76±6	58±8*†‡
T _{re} , °C	37.5±0.1	37.4±0.1	37.5±0.1	37.8±0.1*	39.1±0.1‡	39.1±0.2‡	38.9±0.2‡	39.3±0.1‡
T _{sk} , °C	31.0±0.2	31.1±0.1	31.1±0.2	31.8±0.1*†	35.4±0.2‡	35.5±0.1‡	35.5±0.2‡	35.6±0.1‡
Heart rate, beats/min	102±5	93±4	97±4	113±2*	158±61‡	158±5‡	152±61‡	162±31‡
Body weight, kg	73.2±2.3	72.7±2.7	73.2±2.7	71.4±2.3*	71.8±2.3	71.4±2.7	71.8±2.7	70±2.3*†‡

Values are means ± SE; *n* = 8. T_{re}, rectal temperature; T_{sk}, skin temperature. *Difference from 0.45% IV and 0.9% IV values (*P* < 0.05). †Difference from 0.45% Oral value (*P* < 0.05). ‡Difference from corresponding Pre-Ex value (*P* < 0.05).

saline resulted in a significant rise in plasma sodium, Osm, and plasma AVP. Differences in the findings of our study and that of Thompson et al. may be due to time of observation, as Thompson et al. observed responses immediately following IV infusion and our observations were following 75 min. Restoration of PV was achieved in the present study by IV solutions with differing [Na⁺]. Both IV [Na⁺] equally blunted the response of these fluid regulatory factors with suppression of osmotic and volemic stimuli reducing fluid regulatory responses compared with the NF trial.

ALD and %ΔPV responses. The observation that plasma ALD was not different among the Rehy treatments is likely due to the fact that there were no differences in PV shifts because of the consistent volume infused in the treatments (~1.8 liters). Changes in circulating volume are one of the primary influences on ALD secretion (20). These findings agree with observations in subjects receiving either water or an electrolyte solution before or during exercise in a warm environment, where plasma renin and ALD responses were reduced equally in both treatments (12, 13). Plasma [ALD] also are affected by circadian variation and dietary sodium or potassium intake (7, 17, 28). However, in the present study, experimental testing occurred at the same time of day for each trial, and dietary sodium and potassium intake were similar for 3 days before each treatment.

Oral vs. IV Rehy. Following Rehy, the absolute change in AVP was not different between the 0.45% IV and Oral treatments. In addition, there was no difference observed in plasma Osm between these two treatments, which has been demonstrated to be the major stimulus for AVP release (18). The 0.45% Oral Pre-Ex [Na⁺], however, was significantly lower compared with Pre-Ex values in the NF trial and 0.9% IV treatment. It is possible that this difference may have been due to limitations imposed by gastric emptying (7) and transit compared with the IV treatments. PV has also been shown to be another factor influencing AVP and ALD (5, 6, 18). In the present study, the %ΔPV relative to Pre-Dehy at the Post-Dehy, Pre-Ex, and Post-Ex time points were not different among the Rehy treatments. Given the greater time required for fluid to empty the gut, it might be expected that the %ΔPV would be different between the 0.45% IV and Oral treatments. However, by 75 min following Rehy, restoration of PV was not different between the 0.45% IV and Oral treatments. It is possible that fluid in the 0.45% Oral treatment may have been absorbed across the intestinal lumen, while the sodium may have taken longer to transport into the vasculature, which would explain the lower Pre-Ex [Na⁺] in the 0.45% Oral treatment but the lack of difference in %ΔPV among the

treatments. Differences in [AVP] between the 0.45% IV and Oral treatments may also have been expected due to residual effects of the oropharyngeal reflex. In humans, oropharyngeal receptors have been implicated in the rapid, transient fall in plasma AVP due to drinking (25). In the present study, this effect may have been too late to be observed 75 min following oral Rehy.

Heat tolerance and performance. Previous work (21) has shown that, immediately following exercise, individuals will rehydrate ~60% of the fluid lost. In the present study, subjects did perform exercise in the heat with a ~2% body weight loss following Rehy. We hypothesized that there would be a greater fluid retention with the 0.9% IV treatment compared with the 0.45% IV or Oral treatments, which would impart an advantage when performing exercise in the heat while hypohydrated. However, contrary to this hypothesis, we did not observe greater fluid retention or a difference in exercise performance between the 0.9% and 0.45% IV or Oral saline treatments, as plasma AVP was similar pre-Ex (subsequent to Rehy) and Post-Ex in the heat. Cardiovascular and thermoregulatory strain (elevated heart rate and core temperature) and exercise performance are related to decreased blood volume and increased plasma Osm (18), which were not different in the two IV saline experiments. It is not surprising that the exercise performance time in NF was 24% lower, as the subjects in that trial were twice as dehydrated vs. Rehy trials. The lack of difference between the 0.45% and 0.9% IV treatments for any of the measured variables suggests that the 0.9% IV treatment did not offer any cardiovascular, thermoregulatory, or performance advantages compared with the 0.45% IV tests. It is possible that fluid of greater tonicity than 0.9% saline could result in greater fluid retention and provide physiological and performance advantages.

In summary, the results of the present investigation demonstrate that Rehy with 0.9% IV saline did elevate plasma sodium, Osm, and [AVP] concentrations above levels observed following 0.45% IV Rehy. However, while Rehy with fluid containing sodium has been shown to result in greater restoration of PV compared with water (20), it appears that the greater tonicity of the 0.9% fluid did not result in greater restoration of PV, enhanced heat tolerance, or diminished physiological strain during subsequent exercise in the heat compared with 0.45% saline fluid.

ACKNOWLEDGMENTS

The authors thank the subjects who donated time and effort to participate in this study. The authors also thank Marie Kenefick, Mike Whittlesey, Stavros Kavouras, Doug Casa, Dane McFarland, and Dean Aresco for technical

support. Lastly, the authors thank Michael N. Sawka and Samuel N. Cheuvront for editorial assistance.

GRANTS

This work was supported in part by a grant from the Proctor and Gamble Company and a grant from the University of Connecticut Research Foundation.

DISCLAIMER

The views, opinions, and/or findings in this report are those of the authors and should not be construed as official Department of the Army position, policy, or decision, unless so designated by other official designation. All experiments were carried out in accordance with state and federal guidelines.

REFERENCES

1. **Armstrong LE, Maresh CM, Castellani JW, Bergeron MF, Kenefick RW, LaGasse KE, Riebe D.** Urinary indices of hydration status. *Int J Sport Nutr* 4: 265–279, 1994.
2. **Brandenberger G, Candas V, Follenius M, Libert JP, Kahn JM.** Vascular fluid shifts and endocrine responses to exercise in the heat: effect of rehydration. *Eur J Appl Physiol* 55: 123–129, 1986.
3. **Brozek J, Grande F, Anderson JT, Keys A.** Densitometric analysis of body composition: revision of some quantitative assumptions. *Ann NY Acad Sci* 110: 113–140, 1963.
4. **Candas V, Libert JP, Brandenberger G, Sagot JC, Amoros C, Kahn JM.** Hydration during exercise; effects on thermal and cardiovascular adjustments. *Eur J Appl Physiol Occup Physiol* 55: 113–122, 1986.
5. **Convertino VA, Brock PJ, Keil LC, Bernauer EM, Greenleaf JE.** Exercise training induced hypervolemia: role of plasma albumin, renin, and vasopressin. *J Appl Physiol* 48: 665–669, 1980.
6. **Convertino VA, Keil LC, Bernauer EM, Greenleaf JE.** Plasma volume, osmolality, vasopressin, and renin activity during graded exercise in man. *J Appl Physiol* 50: 123–128, 1981.
7. **Costill DL.** Gastric emptying of fluids during exercise. In: *Perspectives in Exercise Science and Sports Medicine. Fluid Homeostasis During Exercise*, edited by Gisolfi CV and Lamb Dr. Carmel, IN: Brown and Benchmark, 1990, vol. 3, p. 110–114.
8. **Costill DL, Branam G, Fink W, Nelson R.** Exercise induced sodium conservation: changes in plasma renin and aldosterone. *Med Sci Sports Exerc* 8: 209–213, 1976.
9. **Costill DL, Fox EL.** Energetics of marathon running. *Med Sci Sports Exerc* 1: 81–86, 1969.
- 9a. **Dill DB, Costill DL.** Calculation of percentage changes in volumes of blood, plasma, and red cells in dehydration. *J Appl Physiol* 37: 247–248, 1974.
10. **Follenius M, Candas V, Bothorel B, Brandenberger G.** Effect of rehydration on atrial natriuretic peptide release during exercise in the heat. *J Appl Physiol* 66: 2516–2521, 1989.
11. **Fortney SM, Nadel ER, Wenger CB, Bove JR.** Effect of acute alterations of blood volume on circulatory performance in humans. *J Appl Physiol* 50: 292–298, 1981.
12. **Francesconi RP, Sawka MN, Pandolf KB.** Hypohydration and heat acclimation: plasma renin and aldosterone during exercise. *J Appl Physiol* 55: 1790–1794, 1983.
13. **Francis KT, MacGregor R.** Effect of exercise in the heat on plasma renin and aldosterone with either water or a potassium-rich electrolyte solution. *Aviat Space Environ Med* 49: 461–465, 1978.
14. **Katch FI, Katch VL.** Measurement and prediction errors in body composition assessment and the search for the perfect prediction equation. *Res Q Exerc Sport* 51: 249–260, 1980.
15. **Kenefick RW, Maresh CM, Armstrong LE, Castellani JW, Riebe D, Echegaray ME, Kavourous SA.** Plasma vasopressin and aldosterone responses to oral and intravenous saline rehydration. *J Appl Physiol* 89: 2117–2122, 2000.
16. **Maresh CM, Judelson DA.** Alteration in arginine vasopressin with exercise, environmental stress and other modifying factors. In: *The Endocrine System in Sports and Exercise*, edited by Kraemer WJ and Rogol AD. Malden, MA: Blackwell, 2005, p. 487–498.
17. **Metzler CH, Thrasher TN, Keil LC, Ramsay DJ.** Endocrine mechanisms regulating sodium excretion during water deprivation in dogs. *Am J Physiol Regul Integr Comp Physiol* 251: R560–R568, 1986.
18. **Moses AM, Miller M, Streeten DHP.** Quantitative influence of blood volume expansion on the osmotic threshold for vasopressin release. *J Clin Endocrinol Metab* 27: 655–662, 1967.
19. **Nielsen B.** Temperature regulation; effects of sweat loss during prolonged exercise. *Acta Physiol Scand* 128: 105–109, 1986.
20. **Nose H, Mack GW, Shi X, Nadel ER.** Involvement of sodium retention hormones during rehydration in humans. *J Appl Physiol* 65: 332–336, 1988.
21. **Nose H, Mack GW, Shi H, Nadel ER.** Role of osmolality and plasma volume during rehydration in humans. *J Appl Physiol* 69: 609–616, 1990.
22. **Ramanathan NL.** A new weighting system for mean surface temperature of the human body. *J Appl Physiol* 19: 531–533, 1964.
23. **Sawka MN, Francesconi RP, Pimental NA, Pandolf KB.** Hydration and vascular fluid shifts during exercise in the heat. *J Appl Physiol* 56: 91–96, 1984.
24. **Sawka MN, Young AJ, Francesconi RP, Muza SR, Pandolf KB.** Thermoregulatory and blood responses during exercise at graded hypohydration levels. *J Appl Physiol* 59: 1394–1401, 1985.
25. **Seckl JR, Williams TDM, Lightman SL.** Oral and hypertonic saline causes transient fall of vasopressin in humans. *Am J Physiol Regul Integr Comp Physiol* 251: R214–R217, 1986.
26. **Share L, Claybaugh JR.** Regulation of body fluids. *Annu Rev Physiol* 34: 235–260, 1972.
27. **Thompson CJ, Burd JM, Baylis PH.** Acute suppression of plasma vasopressin and thirst after drinking in hypernatremic humans. *Am J Physiol Regul Integr Comp Physiol* 252: R1138–R1142, 1987.
28. **Wade CE, Claybaugh JR.** Plasma renin activity, vasopressin concentration, and urinary excretory responses to exercise in men. *J Appl Physiol* 49: 930–936, 1980.
29. **Wilmore JH, Vodak PA, Parr RB, Girandola RN, Billing JE.** Further simplification of a method for determination of residual lung volume. *Med Sci Sports Exerc* 12: 216–218, 1980.